

Microbiology

Dental Caries

Dental caries is a infectious microbiologic disease of the teeth that results in localized dissolution and destruction of calcified tissues, it as a **"microbial disease"** of the calcify. tissues of the teeth, characterized by demineralization of the inorganic portion and destruction of the organic substance of the tooth.

carious lesion only occur under mass of bacteria capable of producing a sufficiently acidic environment to demineralize the tooth structure . A gelatinous mass of bacteria adhering to the tooth surface is termed **dental plaque** .the plaque bacteria metabolize refined carbohydrate for energy and produce organic acids as a by- product. acids produced may then cause a carious lesion by dissolution of the tooth crystalline structures. Caries lesion progress as a series of exacerbations and remissions as the PH at the tooth surface varies with changes in plaque metabolism .

Exacerbations of caries activity are characterized by periods of high bacterial metabolic activity and low PH in the plaque near the tooth surface. During intervening episodes when few carbohydrates are available. there is little bacterial metabolic activity and the PH rises near the surface of the tooth..Saliva contains high concentration of calcium and phosphate ions in solution which serve as a supply of raw materials of the remineralization process .

Tooth decay, which is also called dental cavities or dental caries, is the destruction of the outer surface (enamel) of a tooth. Decay results from the action of bacteria that live in plaque, which is a sticky, whitish film formed by a protein in saliva (mucin) and sugary substances in the mouth. The plaque bacteria sticking to tooth enamel use the sugar and starch from food particles in the mouth to produce acid.

A disease in which the mineralized tissues of the tooth undergo progressive destruction from the surface of the tooth. It is caused by bacteria that colonize the tooth surface and, under certain conditions, produce sufficient acids to demineralize the enamel covering of the tooth crown or the cemented covering the root, and then the underlying dentin. As the destruction of the dentin progresses, along with breakdown of the organic components, the bacteria invade the dead tissue and enter the pulp chamber. The pulpal tissue becomes infected and the typical toothache may ensue. The infection can ultimately destroy the pulpal tissue and

extend through the apical openings of the roots and into the surrounding periodontal tissues.

The sites of caries development have been correlated with the presence of dental plaque containing one of a number of mutans streptococci—notably in humans. *Streptococcus mutans* and *S. sobrinus*. These microorganisms are cariogenic because of their ability to generate a considerable amount of acid as a result of their metabolism of carbohydrates, and to survive in an acid environment. The acid environment is only slowly neutralized since its dilution is hindered by the fact that the acid is within the dental plaque and thereby shielded from the saliva. Each time a carbohydrate-rich substance is ingested, acid is formed. Therefore, the frequency of ingestion and physical consistency of fermentable carbohydrates are important factors in caries formation.

Dental caries can be prevented by making the tooth less susceptible to acid attack, removing cariogenic bacteria from the teeth, and limiting ingestion of cariogenic substrate

Etiology

There are some 200 to 300 species of microorganisms that have different roles in dental caries. Cavities, are very common. They begin with acid on the tooth. The acid is made from the bacteria in dental plaque. The plaque bacteria feed on sugars and starches from the diet and change them into acid. This acid eats into tooth enamel, or the outer layer of the tooth, and dentin, the major part or core of the tooth. The tooth then gradually dissolves and the bacteria move into the damaged tooth structure.

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Causes and Symptoms

Tooth decay requires the simultaneous presence of three factors: plaque bacteria, sugar, and a vulnerable tooth surface. Although several microorganisms found in the mouth can cause tooth decay, the primary disease agent appears to be *Streptococcus mutans*. The simple sugars used by the bacteria are glucose, sucrose, and lactose.

They are converted primarily into lactic acid. When this acid builds up on an unprotected tooth surface, it dissolves the minerals in the enamel, creating holes

and weak spots (cavities). As the decay spreads inward into the middle layer (the dentin), the tooth becomes more sensitive to temperature and touch. When the decay reaches the center of the tooth (the pulp), the resulting inflammation (pulpitis) produces a toothache.

Root caries, which are sometimes described as a category of smooth-surfaces caries, are the third most common type of caries and usually occur when the root surfaces have been exposed due to gingival recession. When the gingiva is healthy, root caries is unlikely to develop because the root surfaces are not as accessible to bacterial plaque. The root surface is more vulnerable to the demineralization process than enamel because cementum begins to demineralize at 6.7 pH, which is higher than enamel's critical pH. Regardless, it is easier to arrest the progression of root caries than enamel caries because roots have a greater reuptake of fluoride than enamel. Root caries are most likely to be found on facial surfaces, then interproximal surfaces, then lingual surfaces. Mandibular molars are the most common location to find root caries, followed by mandibular premolars, maxillary .anteriors, maxillary posteriors, and mandibular anteriors

Lesions on other smooth surfaces of teeth are also possible. Since these occur in all smooth surface areas of enamel except for interproximal areas, these types of caries are easily detected and are associated with high levels of plaque and diets promoting caries formation] Under Black's classification system, caries near the gingiva on the facial or lingual surfaces is designated Class V. Class VI is reserved for caries confined to cusp tips on posterior teeth or incisal edges of anterior teeth

There are four main criteria required for caries formation: a tooth surface (enamel or dentin); caries-causing bacteria; fermentable carbohydrates (such as sucrose); and time. The caries process does not have an inevitable outcome, and different individuals will be susceptible to different degrees depending on the shape of their teeth, oral hygiene habits, and the buffering capacity of their saliva. Dental caries can occur on any surface of a tooth which is exposed to the oral cavity, but not the structures which are retained within the bone

Bacteria : The mouth contains a wide variety of oral bacteria, but only a few specific species of bacteria are believed to cause dental caries: *Streptococcus mutans* and *Lactobacilli* among them. *Lactobacillus acidophilus*, *Actinomyces viscosus*, *Nocardia* spp., and *Streptococcus mutans* are most closely associated with caries, particularly root caries. Bacteria collect around the teeth and gums in a sticky, creamy-coloured mass called plaque, which serves as a biofilm. Some sites collect plaque more commonly than others. The grooves on the biting surfaces of molar and premolar

teeth provide microscopic retention, as does the point of contact between teeth.

Plaque may also collect along the gingiva

Fermentable carbohydrates

Bacteria in a person's mouth convert glucose, fructose, and most commonly sucrose (table sugar) into acids such as lactic acid through a glycolytic process called fermentation. If left in contact with the tooth, these acids may cause demineralization, which is the dissolution of its mineral content. The process is dynamic, however, as demineralization can also occur if the acid is neutralized by saliva or mouthwash. Fluoride toothpaste or dental varnish may aid remineralization. If demineralization continues over time, enough mineral content may be lost so that the soft organic material left behind disintegrates, forming a cavity or hole. The impact such sugars have on the progress of dental caries is called cariogenicity. Sucrose, although a bound glucose and fructose unit, is in fact more cariogenic than a mixture of equal parts of glucose and fructose. This is due to the bacteria utilizing the energy in the saccharide bond between the glucose and fructose subunits

Role in tooth decay

Early colonizers of the tooth surface are mainly *Neisseria* spp. and streptococci, including *S. mutans*. The growth and metabolism of these pioneer species changes local environmental conditions (e.g. Eh, pH, coaggregation, substrate availability), thereby enabling more fastidious organisms to further colonize after them, forming dental plaque. Along with *S. sobrinus*, *S. mutans* plays a major role in tooth decay, metabolizing sucrose to lactic acid. The acidic environment created in the mouth by this process is what causes the highly mineralized tooth enamel to be vulnerable to decay. *S. mutans* is one of a few specialized organisms equipped with receptors that help for better adhesion to the surface of teeth. Sucrose is utilized by *S. mutans* to produce a sticky, extracellular, dextran-based polysaccharide that allows them to cohere to each other forming plaque. *S. mutans* produces dextran via the enzyme dextransucrase (a hexosyltransferase) using sucrose as a substrate in the following reaction:

$$n \text{ sucrose} \rightarrow (\text{glucose})_n + n \text{ fructose}$$

Sucrose is the only sugar that *S. mutans* can use to form this sticky polysaccharide. Conversely, many other sugars—glucose, fructose, lactose—can be digested by *S. mutans*, but they produce lactic acid as an end product. It is the combination of plaque and acid that leads to dental decay. Due to the role the *S. mutans* plays in tooth decay, there have been many attempts to make a vaccine for the organism. So far, such vaccines have not been successful in humans. Recently, proteins involved in the

colonization of teeth by *S. mutans* have been shown to produce antibodies that inhibit the cariogenic process.

***Streptococcus mutans; the main causative agent of dental caries:-**



carious teeth

Dental Caries: A Bacterial Infection

There are two specific groups of bacteria mostly found in the mouth that are responsible for dental caries ;

1. **Mutans streptococci** (*Streptococcus mutans*)

2. **Lactobacilli**

They are found in relatively large numbers
in the dental plaque

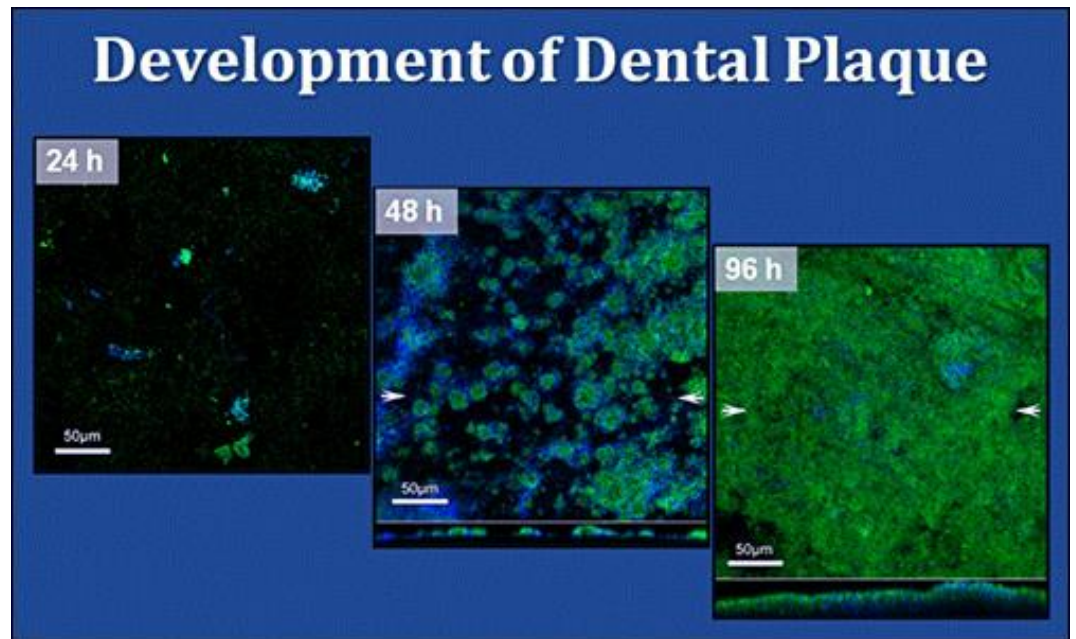
Transmission of Caries Causing Bacteria

Mutans streptococci are transmitted through saliva, most frequently the mother's, to
the infant

When mothers have high counts of mutans streptococci in their mouths, the babies
also have high counts of the same bacteria in their mouth.

Women should be certain their own mouths are healthy.

When the number of caries causing bacteria in the mouth increases, the risk for
developing dental caries also increases.



Microbial Ecology in Health and Disease

Interactions between **mutans streptococci** and **lactobacilli** contribute towards the microbial challenge of dental caries. A comparison was made between numbers of mutans streptococci and lactobacilli in plaque from primary teeth in healthy children and between caries-free teeth and carious lesions in the same mouth of children with caries.

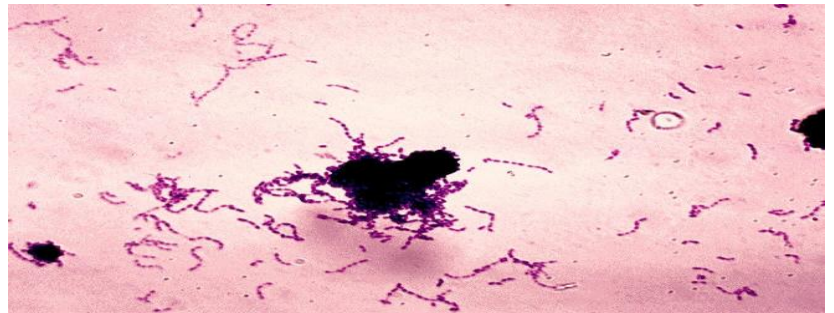
Streptococcus mutans

Description

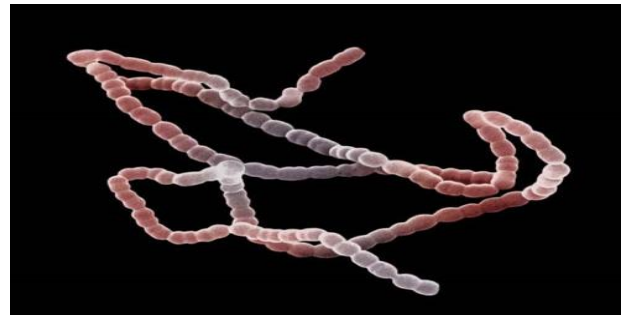
- **Streptococcus mutans** are gram-positive cocci shaped bacteria. These facultative anaerobes are commonly found in the human oral cavity, and is a major contributor of tooth decay. The result of decay can greatly affect the overall health of the individual
- **S. mutans** are mesophilic and grow at temperatures between 18-40 degrees Celsius. **Streptococcus mutans** is a cariogenic microorganism that breaks down sugar for energy and produces an acidic environment, which de-mineralizes the superficial structure of the tooth. The result of the conversion disintegrates the coating of the tooth then later dissolves the Calcium molecule creating a hole. Transmission of **S. mutans** can be found in people of all ages although it is more common for infants and children. The transfer of genotypes are responsible for the transmission of **S. mutans** from mothers to their children although, there is a genotypic variation from one population to the next.

- Streptococcus mutans possesses three virulence factors: water insoluble glycans, acid tolerance, and production of lactic acid
- A toothache is the most common symptom of tooth decay. An infection or irritation of the tooth pulp usually causes the pain.

streptococcus mutans negative stain



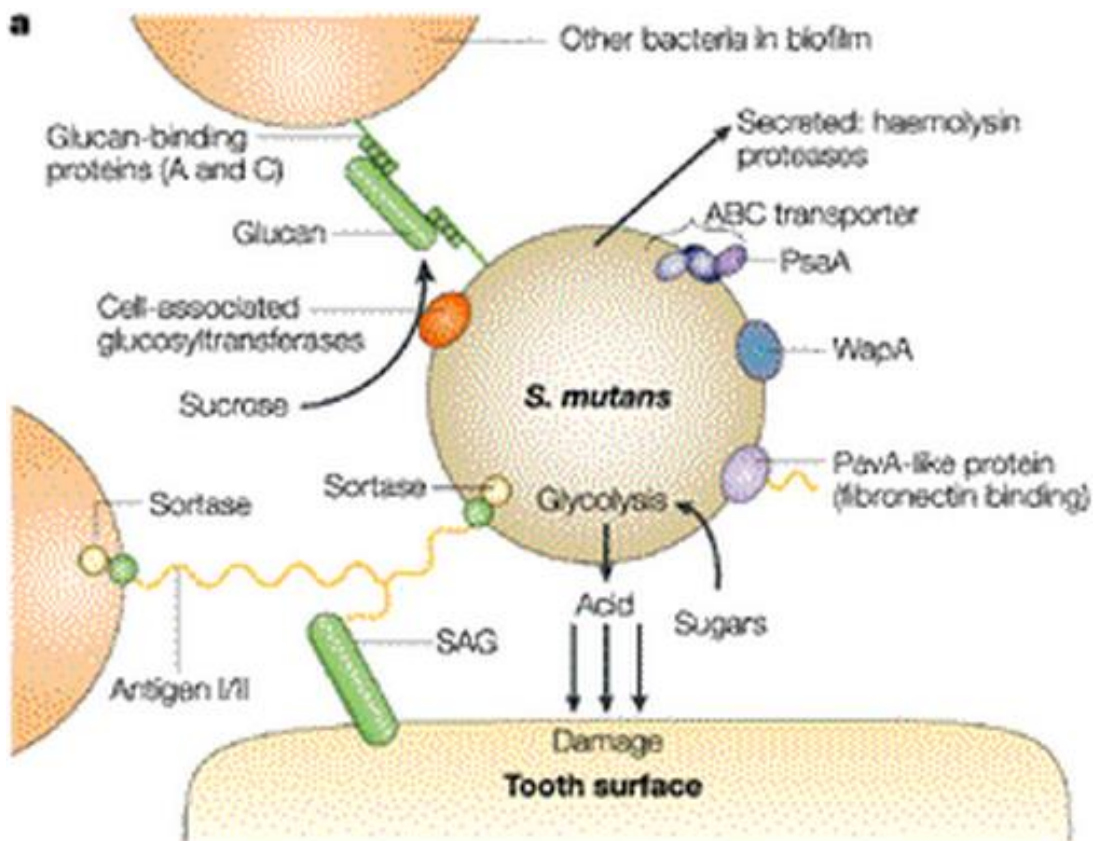
under microscope



Pathogenesis

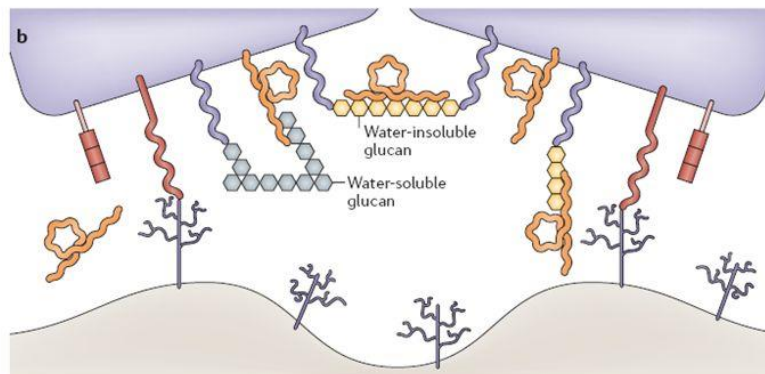
- Streptococcus mutans is considered to be part of the "normal" flora of the human mouth, which is associated with dental caries also known as tooth decay. Dental caries is an infectious and communicable dental disease that affects people of all ages. Streptococcus mutans can be passed from one person to the next via horizontal or vertical transmission. The transmission route of Streptococcus mutans is to colonize itself among human hosts. Children and infants are more prone to the diagnosis of Streptococcus mutans and studies show that many receive it from their primary caregivers

- S. mutans favors hard non-shedding surfaces to establish permanent colonies, which implies that levels of S. mutans are undetected in infants until the primary teeth extravasate. Typically between the ages of one and two, teeth erupt, which allows S. mutans to establish flourishing colonies on teeth, and causes cavities. The indication of cavity formation is the appearance of detectable levels of bacteria on teeth.



The molecular pathogenesis of dental caries associated with mutans streptococci

The molecular pathogenesis of dental caries associated with mutans streptococci



b | Accumulation of mutans streptococci on tooth surfaces in the presence of sucrose. In the presence of sucrose, GTFs synthesize extracellular glucans from glucose (after the breakdown of sucrose into glucose and fructose), and this is thought to be the second event in the formation of dental plaque. The mutans streptococcal protein GBP is a receptor-like protein that is distinct from GTFs, and it specifically binds glucans. GTFs themselves also have a glucan-binding domain and can therefore also function as receptors for glucans. So, mutans streptococci bind pre-formed glucans through GBP and GTFs, and this gives rise to aggregates of mutans streptococci.

Colonization

The growth and metabolism of *S. mutans* changes the environmental conditions of the oral flora, which enables fastidious organisms to colonize and causes the formation of dental plaque. *Streptococcus mutans* is a specialized microorganism equipped with receptors that allow it to adhere to the surface of the tooth creating a slimy environment. After adherence to the tooth, *S. mutans* begin to divide and produce microcolonies within the slime layer to construct a biofilm. *S. mutans* begin to grow and synthesize dextran with the enzyme dextransucrase.

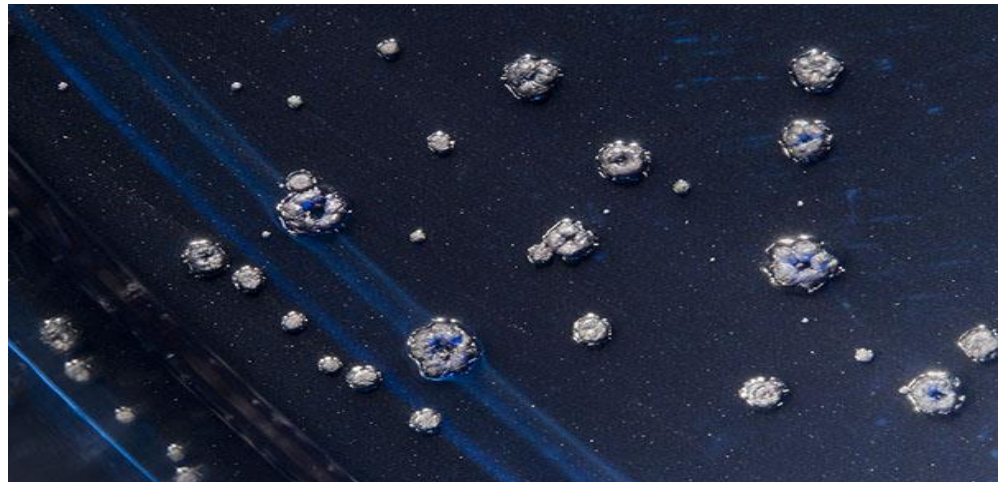
Dextran contains a capsule that binds to the enamel and forms a biofilm that consists of 300-500 bacterial cells. The biofilm metabolizes *S. mutans* and is able to cleave sucrose with the enzyme Glucansucrase in the reaction:



With these products fructose is fermented as an energy source for bacterial growth while glucose is polymerized to an extracellular dextran polymer. This is important because the polymer cements *S. mutans* at the enamel and becomes the matrix of dental plaque. The dextrin can also depolymerize to glucose and use the monosaccharide as a carbon source, which produces lactic acid in the biofilm by

decalcifying the enamel, and leads to dental caries. The combination of acid and plaque results in the causative agent of decay

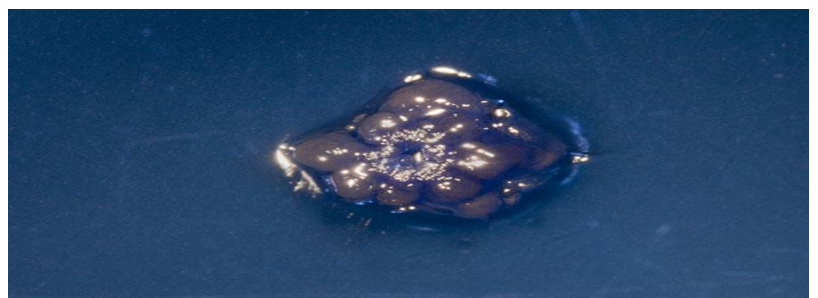
Streptococcus mutans



Streptococcus mutans colonies



. | Streptococcus mutans colony.



Infectious Dose

Bacteria are the most numerous microbes in the mouth. It has been estimated that there are over 100 million in every milliliter of saliva from more than 600 different species

In order to decrease the bacterial challenge, it is encouraged for people to have 10,000 CFU per ml of saliva *Streptococcus mutans* in their mouth.

Colony Forming Units CFU *S. mutans* /ml saliva

class 0-1	<100,000
class 2	100,000< CFU/ml <1,000,000
class 3	>1,000,000 CFU/ml

In the table, class 0-3 is referring to how many *Streptococcus mutans* reside in the mouth with class 0-1 acting as best case with good oral hygiene, while class 3 acts as the worst case

Virulence Factors

There are three virulence factors that are associated with the **cariogenicity** of *Streptococcus mutans*.

The first property is dependent on the synthesis of water-soluble glycans from the disaccharide sucrose.

Next, *S. mutans* possesses the ability to become more acid tolerant. This leads to the third characteristic, which displays a production of lactic acid from dietary sugars



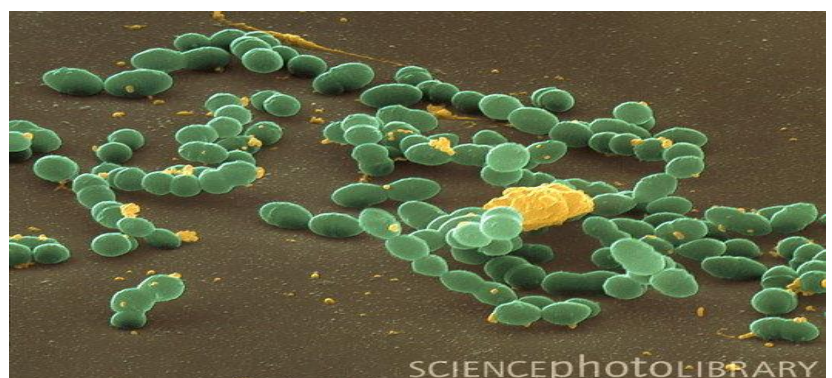
Streptococcus mutans is the major pathogen of dental caries due to the ability to adhere and accumulate to the tooth surface. The Innate Immune Response and the Adaptive Immune Response are two fundamental aspects of the immune system that respond to infections such as dental caries. *Streptococcus mutans* enters the host through the oral route. The mucosal tissue associated with the exocrine glands and saliva contributes to the production of the oral cavity.

This is because; these regions contain cells responsible for antigen internalization and antibodies specific to oral bacteria. The secretory antibodies that protect against dental caries are **IgA** and **IgG**.

The Macrophages of the innate immune response are phagocytic cells that can internalize and kill bacteria by several mechanisms of internalization, which includes macropinocytosis and phagocytosis. If *Streptococcus mutans* initiates dental caries after the teeth have erupted either the host did not undergo the effective immune response of secretory **IgA** played a small role in preventing colonization and subsequent plaque development.

Prevention

It is a fact that every human has bacteria already existing in their mouth. The only prevention is to lessen the impact of the fermentation by product lactic acid. Brushing, flossing and reducing the intake of refined and processed sugars can accomplish this. These processed sugars come in the form of sweets, colas and processed powdered sweeteners. Studies have shown that rinsing one's mouth out with water can greatly reduce the probability of cavities after the consumption of these sugars. It is almost impossible to completely reduce the effects of *S. mutans* but there is an option to control it.



Signs and Symptoms

♦ *Streptococcus mutans* can be identified by a chalky white spot on the surface of the tooth indicating an area of demineralization of enamel, which is commonly referred to as a carious lesion. A carious lesion is the earliest diagnosis of tooth decay formation. As the lesion further demineralizes, it can turn brown and will eventually result in a cavity. Before the formation of the cavity, the process is reversible, but once the *Streptococcus mutans* forms the cavity, the tooth structure is lost and cannot be regenerated.

A lesion that appears shiny and dark brown suggests that a lesion was once present but the demineralization has stopped leaving a stain. As the enamel and dentin are destroyed, the cavity becomes more noticeable.

The effected area of the tooth changes color and becomes sensitive. When the decay passes through the enamel, the dentin tubules allow passages to the nerves making the tooth exposed, which results in pain that temporarily worsens with exposure to heat, cold, sweet food or drinks. If the tooth is weakened by substantial amounts of internal decay a fracture may occur under normal chewing forces. Pain will arise more constant when the decay has progressed enough to where the bacteria can overwhelm the pulp tissue that is located in the center of the tooth.

In addition to cavity formation of the bacterial strain *Streptococcus mutans*, dental caries can also cause bad breath and foul tastes, and gum disease. In highly progressed states, the infection can spread to the soft tissue from the tooth. Life-threatening complications may arise such as cavernous sinus thrombosis and Ludwig's angina

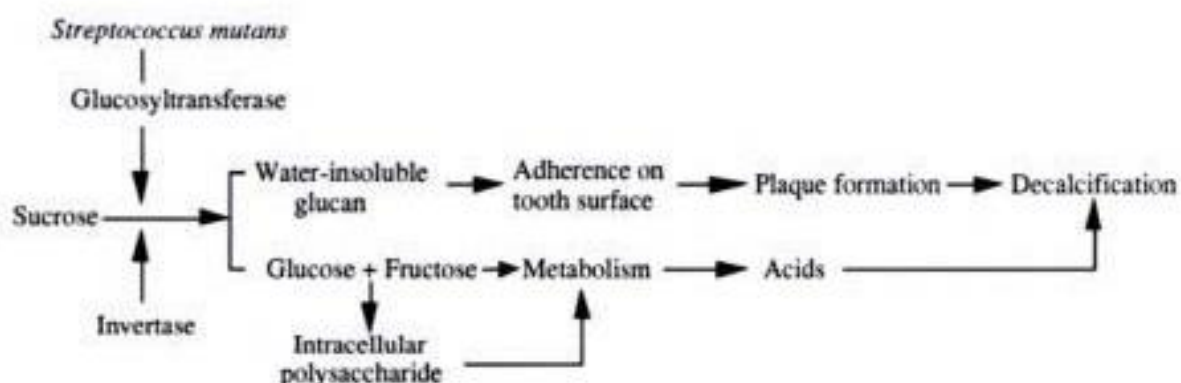


Figure 1. Proposed mechanism of dental caries formation.

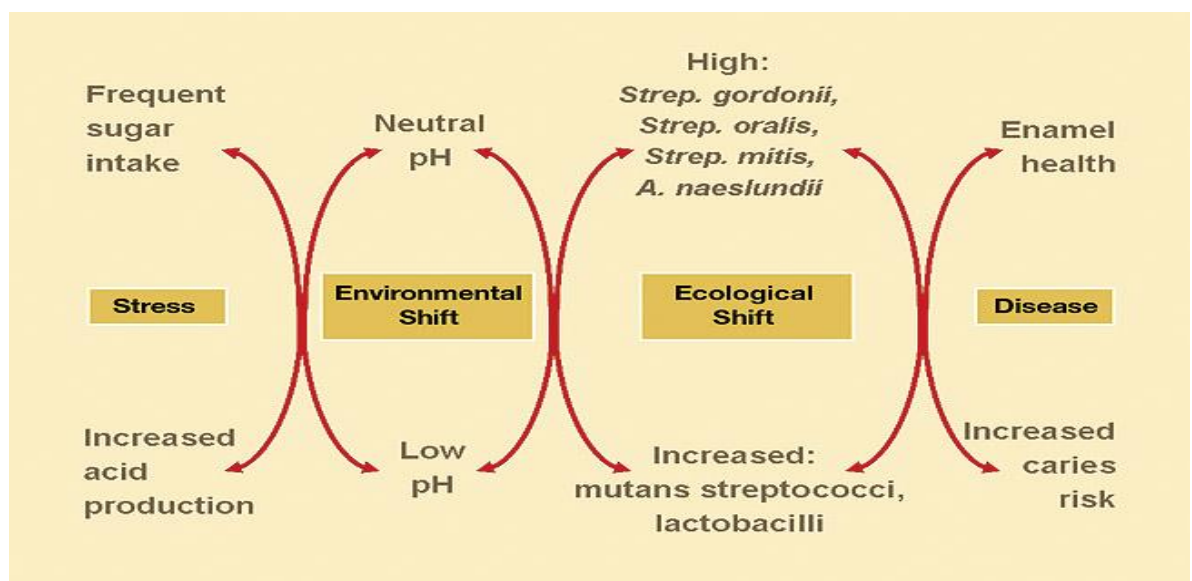
Streptococcus mutans and *Streptococcus sobrinus* are two microorganisms that reside in the mouth of a human.

S. mutans is carried by everyone while *S. sobrinus* is carried between 8 and 35 % of people in different countries. These different bacterial strains can be distinguished by laboratory tests although the procedure is expensive and time consuming; therefore, it is not always practicable to identify the species in a large epidemiological scale.

Researchers not invented a selective media that would allow one to look at the presence of a single species in saliva samples. Because of this, researchers have combined the two species together as mutans streptococci also known as (MS). **Since** *Streptococcus mutans* is more prevalent than *Streptococcus sobrinus* most of the isolates will solely be *S. mutans*. Mutans streptococci can be cultured in a laboratory by taking a sample of the microorganisms in the mouth.

The microbiologist can then use two types of selective media that are widely used to isolate caries-related *Streptococcus* on Mitis- Salivarius agar and TYC agar in which antibiotic Bacitracin is added.

These types of media suppress the growth of most microorganisms but allows the growth of *Streptococcus mutans* and *Streptococcus sobrinus*. The inclusion of sucrose leads to the formation of glucans and the appearance of colony formation to aid identification. Diagnostic kits used in dental clinics are based on similar selective media, but it is important to note that they are measuring for total MS, not just *S. mutans*.



Treatment

Treatment for tooth decay depends on how severe the decay is. One may simply be able to reverse slight tooth decay by the application of **fluoride**. When a processed sugar is consumed, the bacteria from the mouth combine with the sugar to produce an acid. This acid can erode the tooth enamel and damage teeth. Fluoride is used to protect teeth from the demineralization caused by the acid. Fluoride does this by accumulating the demineralized areas of the tooth and strengthening the enamel in a process called remineralization. Fluoride is efficient in preventing tooth decay and making teeth stronger however, it is much less effective if a cavity has already formed

To fix cavities caused by the bacterial strain *Streptococcus mutans* a dentist will need to adequately perform either a composite filling or an amalgam filling- two of the most common fillings used today. In both types of fillings, the dentist will remove the decayed portion of the tooth and fill in the area of the tooth where the decayed area once resided.

